Validation of mule deer body composition using *in vivo* and post-mortem indices of nutritional condition

Thomas R. Stephenson, Vernon C. Bleich, Becky M. Pierce, and Gerald P. Mulcahy

Abstract Measuring fat dynamics is extremely important in understanding the nutritional ecology of cervids. Estimates of body composition provide insight into an animal's energetic state, potential for reproduction and survival, and quality of habitat they occupy. We validated accuracy of *in vivo* and post-mortem indices to predict total body fat in mule deer (*Odocoileus hemionus*). Rump fat thickness measured using ultrasonography was related linearly to ingesta-free body fat, but disappeared at ~5.6% body fat. Loin muscle thickness determined by ultrasonography and condition scores exhibited potential to quantify nutritional reserves when rump fat has been depleted. Whole body mass did not predict body fat in adult females, but was a good predictor of ingesta-free lean body mass. Kidney fat mass and kidney fat index were moderate predictors of total body fat, but the relationship was curvilinear. Of the post-mortem indices we tested, the Kistner Score exhibited the strongest linear relationship to ingesta-free body fat. We suggest that using an accurate *in vivo* method to assess nutritional reserves reveals much about past nutritional history and future productivity of individuals within a population, especially when determined repeatedly in radiocollared animals.

Key words body composition, body condition, fat dynamics, mule deer, nutrition, *Odocoileus hemionus*, ultrasonography

Body condition in North American deer (*Odocoileus* spp.) has been assessed using marrow fat (Cheatum 1949, Torbit et al. 1988), kidney fat (Finger et al. 1981, Anderson et al. 1990), back fat (Anderson et al. 1972), a combination of kidney and marrow fat (Ransom 1965, Connolly 1981), visual scores (Kistner et al. 1980), and water dilution (Torbit 1985*a*). Full utility of these indices requires that their relationship to total body fat be determined. Finger (1981) and Torbit et al. (1985*a*, 1988) related some of the above indices to total body fat, but further validation is needed, particularly for indices that may be used with live deer. Recently, ultrasonography was used to predict total body fat *in vivo* for moose (*Alces alces*) and elk (*Cervus elaphus*; Stephenson et al. 1998 and Cook et al. 2001*a*). *In vivo* methods to determine body composition offer several advantages over the post-mortem indices that have been the focus of previous efforts. In particular, they lend themselves to

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use in repeated measures sampling designs that are more powerful, thus requiring smaller sample sizes, and they are nonlethal. Body composition of radio-collared individuals may be determined during recaptures and related to numerous life history traits and environmental conditions.

Franzmann (1977, 1985) suggested monitoring the body condition of an animal to assess the quality of its environment. Nutritional condition has been defined as the state of body components controlled by nutrition and which, in turn, influence an animal's future fitness (Harder and Kirkpatrick 1994, Grubb 1995, Saltz et al. 1995). The nutrient reserves of an animal are determined by a multitude of processes interacting to produce a continuously varying state. Until thresholds are attained, excess intake of energy and protein result in increased deposition of fat and muscle. Conversely, submaintenance diets produce deficiencies that are countered by using body reserves. In addition to diet, body condition is controlled through behaviors that result in expenditure of reserves, specifically activity, and the costs of thermoregulation and reproduction that use significant resources. Furthermore, disease and stress also may operate to limit or reduce physical condition of individuals.

In modeling mule deer (*Odocoileus hemionus*) survival, accurate estimation of mule deer body fat was essential, given the sensitivity of starvation estimates to prewinter fatness (Hobbs 1989). Estimates of body condition have been used to identify thresholds of fecundity in many ungulates (Cameron and Ver Hoef 1994, Testa and Adams 1998, Cook 2000, Keech et al. 2000). Recently, Keech et al. (2000) determined that maternal condition was related to numerous reproductive parameters in Alaskan moose.

Our objectives were to 1) validate the application of ultrasonography to predict body fat in live mule deer, 2) assess the utility of existing postmortem indices to predict body fat in mule deer, and 3) compare equations relating ingesta-free body fat to rump fat thickness for mule deer with equations developed for moose.

Study area

We conducted our study at Round Valley in eastern California (37°N, 118°W) at the base of the east side of the Sierra Nevada. Vegetation on the mule deer winter range was composed of a mixture of sagebrush (*Artemesia tridentata*), bitterbrush (*Purshia tridentata*), rabbitbrush (*Chrysothamnus nauseosum*), blackbrush (*Coleogyne ramosissima*), and mormon tea (*Ephedra nevadensis*). Associated riparian areas supported willow (*Salix* spp.), *Rosa* spp., and water birch (*Betula occidentalis*). Kucera (1997) described the study area in detail.

Mule deer on the Round Valley (Inyo County, California) winter range using characteristic bitterbrush/sagebrush habitat. The ultrasound technique that we validated was used to estimate overwinter nutritional condition of individuals in this population. Photo by Tom Stephenson.

Methods

Thirteen adult (2-7-yr) female mule deer in Round Valley, Inyo County, California, were shot during March 1996 and 1997 to evaluate body composition. Upon recovery of dead deer, we positioned animals in sternal recumbency and immediately measured rump fat thickness using an Aloka model 210 portable ultrasound device (Aloka, Inc., Wallingford, Conn.) with a 5-MHz 8-cm linear-array transducer. We measured subcutaneous fat thickness at its thickest point immediately cranial to the cranial process of the tuber ischium (pin bone). We measured maximum fat thickness with electronic calipers to the nearest 0.1 cm. We determined body mass of whole animals, and we processed the remains as described by Stephenson et al. (1998). We determined kidney fat mass and calculated the kidney fat index as described by Riney (1955) and Anderson et al. (1990). We visually estimated amount of fat at 6 indicator sites according to Kistner et al. (1980); however, we did not assign points to body musculature, so our score (Kistner Score) varied between 0 and 90. Field-dressed mass was determined from the eviscerated carcass with hide attached. Fat in the carcass and viscera (excluding ingesta and uterine contents) were determined chemically by ether extract and expressed as a percentage. We calculated percentage ingesta-free body fat by summing the products of each component's (including hide) percentage fat and its respective mass, dividing by ingesta-free body mass, and multiplying by 100. We calculated ingesta-free, lean body mass as ingesta-free body mass less the fat mass.

We used linear regression to determine relationships between fat depots (carcass and viscera) and to develop predictive equations for ingesta-free body fat and ingesta-free lean body mass. Postmortem predictive variables included kidney fat index, kidney fat mass, Kistner Score, and field-dressed mass. We log-transformed curvilinear data when appropriate. We used stepwise regression to generate additional predictive equations of ingesta-free body fat for live animals using multiple dependent variables (fat thickness, body mass, total length, and chest girth). To permit comparison of the predictive equation for deer to that existing for moose (Stephenson et al. 1998), we scaled deer to moose using a scaling factor (2.93) that was the ratio of deer to moose fat thickness. We used an *F*-test to test for differences in slope between the scaled versions of the regression equations to predict ingesta-free body fat from fat thickness in deer and moose.

Additionally, we captured and manually restrained 82 adult female mule deer during March 2000 as part of an ongoing study. We measured maximum fat thickness and thickness of the longissimus dorsi (loin) using ultrasonography. We also scored animals using a subjective condition score developed for elk by Cook (2000) and modified for deer by L. Bender (New Mexico State University, personal communication). To evaluate efficacy of measuring loin thickness to detect changes in body composition as fat thickness disappears, we plotted distribution curves of the variable and used a *t*-test to test for a difference in loin thickness between animals with and without measurable fat thickness. We also regressed condition score on loin thickness.

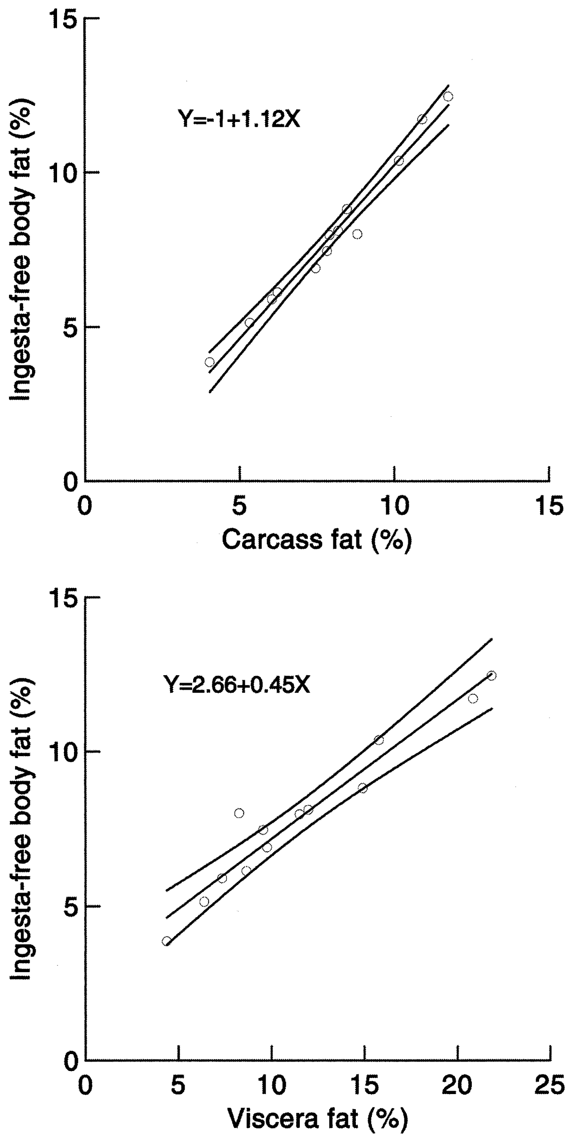


Figure 1. Relationship between carcass fat, viscera fat, and ingesta-free body fat (less concepta) determined by chemical extraction of adult female mule deer carcasses in Inyo County, California, 1996 and 1997. Outer lines represent 95% confidence intervals.

Results

We observed a strong linear relationship between ingesta-free body fat and carcass fat (r^2= 0.98, n=13, P≤0.001, SE=0.4) and viscera fat (r^2= 0.94, n=13, P≤0.001, SE=0.65, Figure 1). Maximum fat thickness, measured using ultrasonography, predicted percentage ingesta-free body fat for values above 5.7% ingesta-free body fat (r^2=0.83, n=13, P ≥0.001, SE=1.07) and mass of ingesta-free body fat (r^2=0.85, n=13, P≤0.001, SE=0.57, Figure 2). Multiple regression that incorporated multiple dependent variables failed to improve predictive equations. Upon scaling the deer fat thickness equation to that of moose for predicting percentage ingesta-free body fat (Figure 3), slopes of the regression

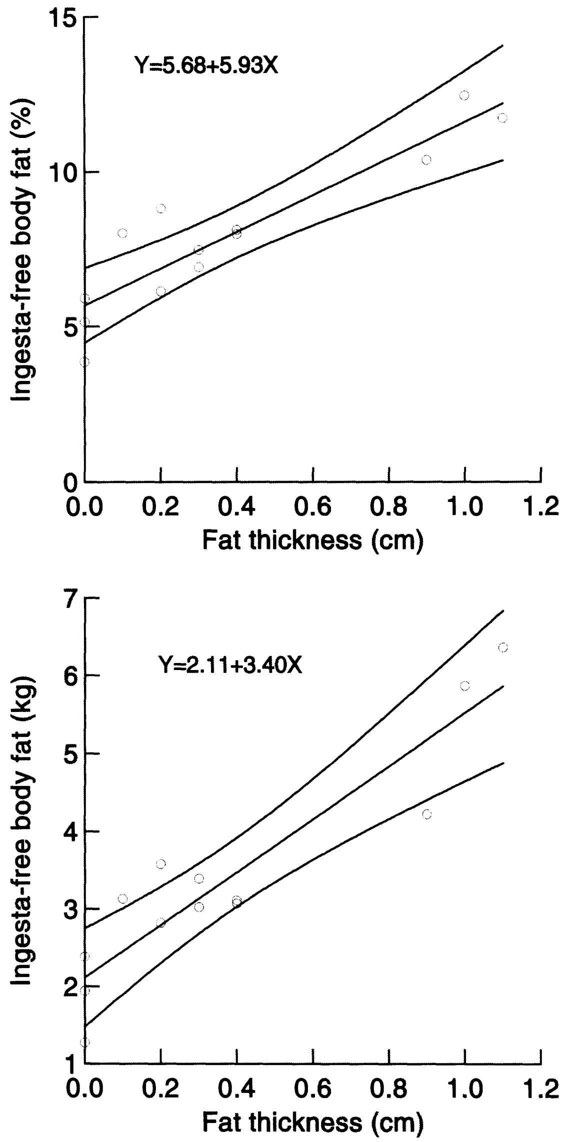


Figure 2. Relationship between maximum rump fat thickness measured using ultrasonography and ingesta-free body fat (less concepta) determined by chemical extraction of adult female mule deer carcasses in Inyo County, California, 1996 and 1997. Outer lines represent 95% confidence intervals.

lines did not differ ($F_{1,18}$=0.007, P=0.94).

Post-mortem indices tested in this study were poorer predictors of ingesta-free body fat (Table 1) than the *in vivo* ultrasonographic maximum fat thickness (Figure 2). Although body mass was a poor predictor of ingesta-free body fat (Table 1), it accurately predicted ingesta-free lean body mass (r^2 =0.81, n=13, $P\leq$0.001, SE=2.27, Figure 4). Field-dressed mass also was a poor predictor of ingesta-free body fat (Table 1). Log-transformed versions of kidney fat index and kidney fat mass were better

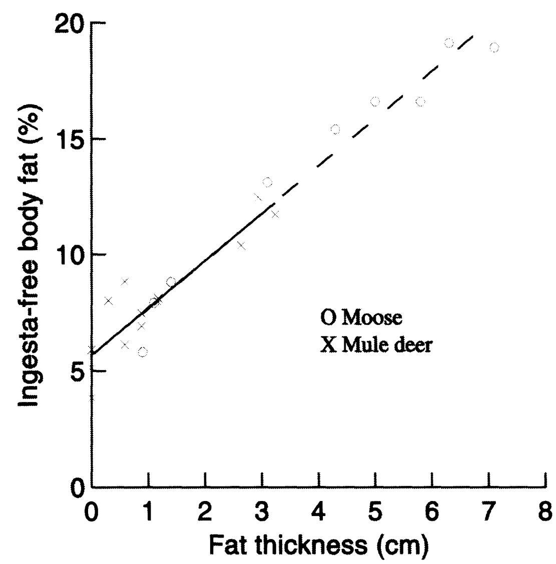


Figure 3. Relationship between maximum rump fat thickness measured using ultrasonography and ingesta-free body fat (less concepta) determined by chemical extraction of adult female mule deer carcasses in Inyo County, California, 1996 and 1997, and moose at the Kenai Moose Research Center, Alaska, 1993–1995. For comparative purposes, mule deer fat thickness was scaled to that of moose (Stephenson et al. 1998).

predictors of ingesta-free body fat than untransformed (Table 1). Kistner Score was the best post-mortem predictor of ingesta-free body fat and exhibited a linear relationship over the range of data tested (Table 1).

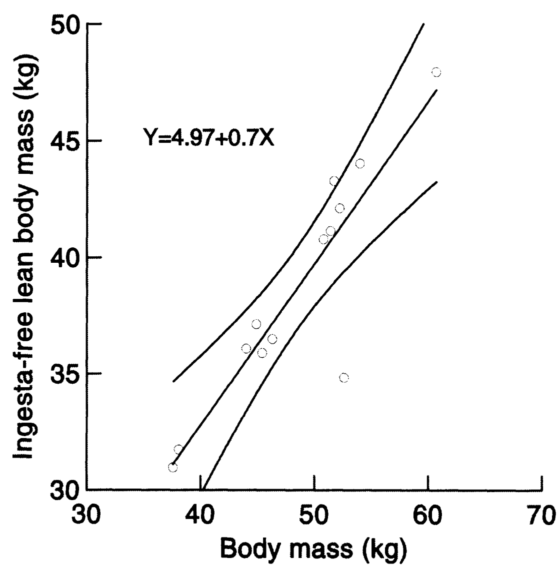


Figure 4. Relationship between body mass and ingesta-free lean body mass in adult female mule deer in Inyo County, California, 1996 and 1997. Outer lines represent 95% confidence intervals.

Table 1. Linear regression equations relating ingesta-free body fat (IFBFAT) to kidney fat index (KFI), kidney fat mass (KFM), Kistner score (KISTSCR, Kistner et al. 1980), field-dressed mass (FDMASS), and body mass for adult female mule deer collected in Inyo County, California, March 1996 and 1997.

Y	*X*	Equation	r^2	*P*	SE	Range
IFBFAT (%)	Mean KFI	$Y = 5.537 + 0.036X$	0.531	0.005	1.784	12.5–163.9%
	Ln mean KFI	$Y = -1.706 + 2.449X$	0.607	0.002	1.633	2.6–5.1
	Mean KFM	$Y = 5.328 + 0.058X$	0.516	0.006	1.813	8.0–100.9 g
	Ln mean KFM	$Y = -1.351 + 2.580X$	0.594	0.002	1.660	2.2–4.6
	KISTSCR	$Y = 0.973 + 0.149X$	0.646	0.001	1.550	25–70
	FDMASS	$Y = 0.82 + 0.25X$	0.29	0.059	2.2	21.9–40.0 kg
	BODYMASS	$Y = -0.70 + 0.18X$	0.22	0.11	2.307	37.6–60.7 kg

Loin thickness differed ($t_{51} = -4.23$, $P \leq 0.001$) between adult females with and without measurable fat thickness (Figure 5). Furthermore, the range of loin thickness in individuals without measurable fat thickness (range = 2.9–4.2 cm) exceeded that of individuals with fat thickness $\geq$0.1 cm (range = 3.4–4.5 cm). We detected a linear relationship between loin thickness and condition score when using deer with fat thickness =0 ($r^2 = 0.17$, $n = 29$, $P = 0.027$, SE = 0.57), but no relationship existed for deer with fat thickness >0 ($r^2 = 0.06$, $n = 24$, $P = 0.2$).

Discussion

Aside from chemical analysis of whole or major portions (carcass, viscera) of mule deer, post-mortem indices of body composition exhibited poorer predictability of total body fat than did rump fat thickness measured using ultrasonography. Insignificant relationships of body mass and field-dressed mass precluded their application for predicting body fat. Because of curvilinearity, log transformations improved the fit of kidney fat index and kidney fat mass to ingesta-free body fat. Limited sample size in this study likely contributed to the reduced strength of our equations, particularly for kidney fat index, compared to those previously published for deer (Finger et al. 1981, Torbit et al. 1988). Anderson et al. (1990) preferred kidney fat mass to kidney fat index because of concern that kidney fat index was biased by annual variation in kidney mass. Even with high coefficients of determination, the nonlinear deposition of kidney fat limits its application over the full range of body fatness. The Kistner Score (Kistner et al. 1980) exhibited the best ability to predict ingesta-free body fat over the broadest range of condition among the post-mortem indices.

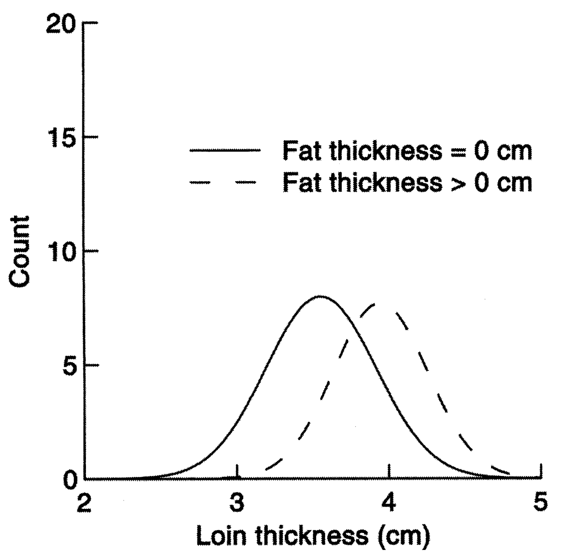


Figure 5. Gaussian distributions of loin thickness measured by ultrasonography in mule deer in Inyo County, California, March 2000. The distributions represent those adult females with no measurable fat thickness (ingesta-free body fat < 5.6%) and those with measurable fat (ingesta-free body fat between 5.6 and 9.8%).

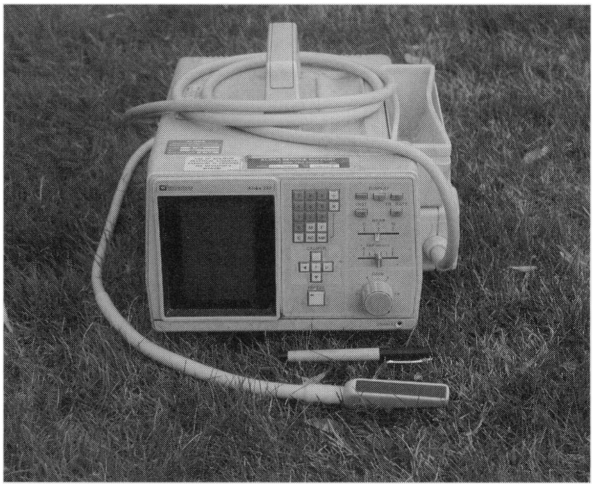
Widespread misinterpretation of patterns of fat catabolism likely has limited the application of indices that rely on measurements of subcutaneous fat reserves. Subcutaneous reserves are not exhausted prior to initiation of mobilization of visceral and marrow deposits. On the contrary, as we observed, the linear fit of carcass and viscera fat (Figure 1) to ingesta-free body fat confirmed that these reserves were mobilized concurrently. Furthermore, carcass and viscera fat appeared to be depleted at values approaching 0% ingesta-free body fat with perhaps carcass fat being exhausted last. The carcass fat component in this study was composed of subcutaneous, intramuscular, and marrow fat. Although subcutaneous fat depots were the first to disappear completely, because most other depots were mobilized concurrently, they were largely depleted when subcutaneous fat was exhausted (Riney 1955). In reindeer (*Rangifer tarandus platyrhynchus*; Reimers and Ringberg 1983) and cattle (Berg and Butterfield 1976), back fat was an accurate predictor of body fat over a wide range. More recently, Stephenson et al. (1998)

and Cook et al. (2001*a*) predicted total body fat with a high degree of accuracy using maximum rump fat thickness in 2 cervid species.

The relationship between total body fat and rump fat thickness in deer and moose exhibited remarkable similarity when scaled to body size (Figure 3). As in moose (Stephenson et al. 1998) and elk (Cook et al. 2001*b*), fat thickness disappeared at ~5.6% ingesta-free body fat in deer, thus limiting application of the technique when an animal was in poorest condition. Consequently, use of an additional measure of condition such as thickness of the longissimus dorsi or a visual condition score was needed to make predictions in the low portion of the range. Because ungulates appear to increase mobilization of muscle (protein reserves) as fat reserves are depleted (Torbit et al. 1985*b*, Cook 2000), detectable changes in muscle thickness are indicative of remaining energy supplies. R. Cook (National Council for Air and Stream Improvement, personal communication) detected a linear relationship between loin thickness and total body fat in elk.

Full application of indices of nutritional condition requires an understanding of their ecological context. In particular, this implies determining the shape of the mathematical relationship between a fat index and total body fat in an animal. Numerous authors (Robbins 1983, Stephenson et al. 1998, Cook et al. 2001*b*) have emphasized the necessity of using predictive equations that are linear to ensure detection of direct relationships along a continuum of condition. Ultimately, we wish to understand the relationship between a nutritional index and an animal's future fitness (Saltz et al. 1995). Body fat and protein reserves are a direct indication of nutritional status and as such are used to make predictions about potential survival and reproduction (Hobbs 1989, Stephenson 1995, Moen et al. 1997). Fat reserves are related directly to the productivity of adult females in ungulate populations (Testa and Adams 1998, Cook 2000, Keech et al. 2000). Numerous examples exist that illustrate the inherent difficulties in applying indices that are often correlated poorly with nutritional status and lack causality when applied in the field (Ruthven et al. 1994, Saltz et al. 1995, Kucera 1997).

Nutritional reserves reflect past dietary quality and energetic costs and predict future survival and reproductive potential. Estimating total body fat, in contrast to simply indexing condition, allows for greater insight into the causal mechanisms that determine population productivity and growth rates because we can quantify energy available for mobilization. We described a procedure that permits rapid determination of total body fat *in vivo*. Indeed, total body fat may be determined repeatedly in marked individuals across seasons and years. Application of this method using a repeated-measures experimental design would reduce sample size requirements and increase the power to detect differences among treatments, years, and locations. The animal histories compiled using repeated captures would eliminate the necessity of inefficient sampling that has been a criticism of other techniques (Saltz et al. 1995) and permit a more comprehensive interpretation of the causal factors that affect condition.



The veterinary ultrasound equipment used during this project is highly portable and may be readily transported and operated in the field. This photo depicts a 5 MHz transducer that is used for measuring fat and muscle thickness. Photo by Tom Stephenson.

Further validation is needed to refine prediction of protein reserves and fat reserves below the point where measurement is limited using fat thickness by ultrasonography (~5.6% ingesta-free body fat). Given variation observed in loin muscle thickness in deer, an approach similar to the LIVEINDEX in elk validated by Cook et al. (2001*a*) appears promising for estimating the entire range of body fatness in mule deer. In addition, further evaluation of condition scoring for estimating body fat in lean animals is warranted (Cook et al. 2001*a*).

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